REMARKS

The Official Action dated September 9, 2005 has been carefully considered. Accordingly, it is believed that the present Amendment places this application in condition for allowance. Reconsideration is respectfully requested.

By the present Amendment, claim 5 is amended to recite that the pH of the cells after addition of the metal salt is equal to or lower than pH 6.8, as set forth in the Examples and at page 3, lines 8-9 of the specification. Claims 21 and 22 are amended to recite that the pH of the cells after the addition of the metal salt is less than or equal to 7, again in accordance with the teachings of the specification at page 3 and in the Examples. Claims 21 and 22 have also been amended to recite the effect of reducing the amount of trisulfides formed in the production of the recombinant growth hormone (claim 21) or recombinant peptide (claim 22), as set forth throughout the specification, and to recite that the production does not include a peptide refolding step after the addition of the metal salt, as set forth at page 2, lines 23-27 (i.e., "not, as earlier suggested, by conversion of the formed trisulfide of growth hormone into the native form") and in Example 6 (wherein the expression is to the periplasm resulting in soluble folded protein without a refolding step). It is believed that these changes do not involve any introduction of new matter, whereby entry is believed to be in order and is respectfully requested.

In the Official Action, claims 5-7, 11-14, 16-19, 21 and 22 were rejected under 35 U.S.C. §103(a) as being unpatentable over the Builder et al U.S. Patent No. 5,663,304. The Examiner again asserted that Builder et al teach a method for production of recombinant peptides comprising fermenting cells to produce recombinant peptides in the presence of metal salt prior to peptide isolation and that the metals facilitate trisulfide oxidation of polypeptides and yield correct refolding of a misfolded polypeptide contained in host cells. In response to Applicants' previous arguments, the Examiner asserted that limitations in the preambles of the claims are not given patentable weight. Additionally, the Examiner asserted that the addition of metal salt taught by Builder et al inherently facilitates the formation of disulfide or trisulfides and Builder et al teach the method to produce recombinant peptides which are correctly folded.

This rejection is traversed and reconsideration is respectfully requested with respect to pending claims 5-7, 11-14, 16, 17, 21 and 22. Applicants submit that the methods defined by

these claims are not suggested by Builder et al and are patentably distinguishable from the teachings of Builder et al.

More particularly, independent claim 21 is directed to a method for the production of recombinant growth hormone, comprising fermenting cells to produce recombinant growth hormone. Independent claim 22 is directed to a method for the production of recombinant peptides, comprising fermenting cells to produce recombinant peptides. Claims 21 and 22 specify that a metal salt is added during or after the fermentation step, prior to growth hormone isolation (claim 21) or peptide isolation (claim 22). Additionally, the pH of the cells after the addition of the metal salt is less than or equal to 7. Both claims 21 and 22 further recite the result of reducing the amount of trisulfides formed in a production of the growth hormone (claim 21) or recombinant peptide (claim 22), and the proviso that the production does not include a peptide refolding step after the addition of the metal salt.

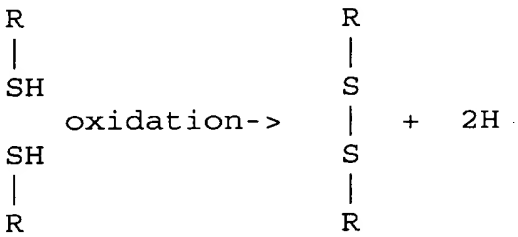
In contrast to the present methods, Builder et al disclose a method of refolding misfolded polypeptide contained in host cells. Builder et al disclose that the "essence" of their invention is utilizing a special buffer containing a minimal concentration of copper or manganese salt to enhance refolding of misfolded polypeptides (column 7, lines 10-12). The special buffer is disclosed as having a pH of 7-12 and comprising about 5-40% (v/v) of an alcoholic or polar aprotic solvent, about 0.2 to 3 M of an alkaline earth, alkali metal or ammonium salt, about 0.1 to 9 M of a chaotropic agent, and about 0.01 to 15 μm of a copper or manganese salt (column 6, lines 42-50).

However, Applicants find no teaching or suggestion by Builder et al relating to a method as recited in claim 21 or claim 22 wherein the amount of trisulfides formed in the production of growth hormone or recombinant peptide is reduced. Particularly, Applicants find no teaching or suggestion by Builder et al of such a method wherein the production does not include a peptide refolding step after the addition of a metal salt, as required by claims 21 and 22. To the contrary, Builder et al disclose the addition of their buffer containing a metal salt to obtain refolding of a misfolded polypeptide, and Builder et al are silent as to the problem of trisulfide formation in recombinant polypeptide production, particularly recombinant growth hormone production, and therefore provide no teaching or suggestion of a solution to this problem.

At page 5 of the Official Action, the Examiner asserts that the addition of a metal salt inherently facilitates the formation of disulfide or trisulfides, which is dependent on the

concentration of metal salt used in the process. Initially, Applicants note that the presently claimed methods reduce the amount of trisulfides formed in the production of the growth hormone or the recombinant peptide, whereby a method facilitating trisulfide production is undesirable.

Moreover, refolding involves breaking incorrect disulfide bonds and allowing correct disulfide bonding (column 2, lines 38-42). Disulfide oxidation as desired by Builder et al is not the same as reducing trisulfide formation during production, prior to isolation. That is, a disulfide bond (S-S bond), also called a disulfide bridge, is a strong covalent bond between two sulfhydryl groups. This bond is important to the folding, structure, and function of proteins. When two amino acids bond to each other through their side chains, they normally do so through a disulfide bond. The particular side chain involved is the sulfhydryl group (-SH). Oxidation of the thiol group yields a disulfide (S-S) bond. The presence of S-S then helps to maintain the tertiary structure of the protein. An amino acid that commonly forms S-S bonds in proteins is cysteine. When two cysteines are bonded by an S-S bond, the resulting molecule between the two protein chains is called cystine. The figure below shows the formation of a disulfide bond. The R on each side group represents the remainder of the amino acid.



Builder et al, teach that metal salts, specifically of copper and manganese (transition metals), facilitate thiol oxidation to form a disulfide bond. The Examiner's extension of this teaching, suggesting that if metal salts facilitate disulfide bond formation, metal salts would also facilitate trisulfide bond formation, is opposite to the present methods which claim reducing trisulfide formation. Therefore, as the present invention is directed towards methods reducing the amount of trisulfide in the production of recombinant peptides, Builder et al teach away from the present invention. As a "useful general rule", references that teach away cannot serve to create a prima facie case of obviousness, *In re Gurley*, 27 F.3d 551, 553

(Fed. Cir. 1994). Accordingly, the present invention, which solves the problem of trisulfide formation in peptides, is non-obvious from the teachings of Builder et al.

At page 5 of the Official Action, the Examiner disregards Applicants' reliance on *In re Gurley, supra*, on the basis that the addition of metal salt to the method inherently facilitates the formation of disulfide or trisulfides. First, Applicants note that inherency and obviousness are entirely different concepts, *In re Reinhart*, 189 U.S.P.Q. 143, 148 (C.C.P.A. 1976), so that any improvements which might inherently result from the Examiner's modification of the teachings of Builder et al to result in the presently claimed methods are irrelevant if those improvements are not suggested by Builder et al. Moreover, as noted above, Applicants' methods result in the reduction of trisulfides formed in the production methods, whereby teachings of Builder et al which inherently facilitate trisulfide production are contrary to the present methods, and Builder et al provide no teaching or suggestion for reducing trisulfide formation in the production of growth hormone or recombinant peptides.

The Examiner also asserted that Builder et al teach a method of producing polypeptides in the presence of an alkali metal salt prior to isolation. To support this assertion the Examiner refers to the fermentation exemplified in the specification where an alkali metal salt is a component of the culture medium (column 23, line 26 - column 27, line 30). However, Builder et al disclose that: "*production of IGF-1 occurred after the phosphate in the medium was depleted*". (column 27, lines 25-26) The source of the phosphate is the alkali metal salt. If the phosphate is being depleted prior to the production of the polypeptide, there can be no teaching or suggestion by Builder et al of the presently claimed methods because the present methods employ metal salt during or after fermentation to produce recombinant peptides with a reduction in the amount of trisulfides.

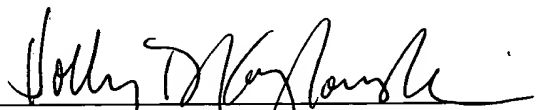
The Examiner has referred to various other teachings in Builder et al as disclosing the use of metal salts. However, as Builder et al teach the use of such salts in a method of refolding a polypeptide, these various disclosures of Builder et al do not teach or suggest the methods defined by claims 21 and 22, wherein a metal salt is added during or after the fermentation step in a production that does not include a peptide refolding step after addition of a metal salt as required by claims 21 and 22. Thus, the teachings of Builder et al do not inherently result in a method as presently claimed, nor do Builder et al provide any teaching or suggestion for modifying the methods disclosed therein to result in such methods.

To establish prima facie obviousness of the claimed invention, all of the claim limitations must be taught or suggested by the prior art, *In re Royka*, 180 U.S.P.Q. 580 (C.C.P.A. 1974). Furthermore, references relied upon to support a rejection under 35 U.S.C. §103 must provide an enabling disclosure, i.e., they must place the claimed invention in the possession of the public, *In re Payne,* 203 U.S.P.Q. 245 (C.C.P.A. 1979). Not only do Applicants find no teaching, suggestion or reference by Builder et al of the methods as defined by claims 21 and 22, Applicants find no teaching, suggestion or reference in Builder et al for modifying the disclosures therein to arrive at the claimed methods. In view of the failure of Builder et al to teach, suggest or recognize the methods as defined by claims 21 and 22, Builder et al do not provide an enabling disclosure of the present methods and therefore do not support a rejection of claims 5-7, 11-14, 15, 17, 21 and 22 under 35 U.S.C. §103.

Finally, Applicants note that claim 3 has not been rejected. Accordingly, an indication of the allowability of claim 3 is respectfully requested.

It is believed the above represents a complete response to the Official Action and places the present application in condition for allowance. Reconsideration and an allowance are requested.

Respectfully submitted,



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